

Claims

1. Procaryotic host cells which are genetically modified for enhanced synthesis of at least one polyketide, wherein said modification comprises incorporation of at least one expression system for producing a protein that catalyzes the production of starter and/or extender units and/or disabling at least one endogenous pathway for catabolism of starter and/or extender units.

2. The cells of claim 1 which are of the genus *Escherichia*, *Streptomyces*, *Bacillus*, *Pseudomonas*, or *Flavobacterium*.

3. The cells of claim 2 which are *E. coli*.

4. The cells of claim 1 which produce a complete polyketide.

5. The cells of claim 3 which produce a complete polyketide.

6. The cells of claim 4 wherein the polyketide is 6-dEB.

7. The cells of claim 1 which do not produce polyketide in the absence of genetic modification, and wherein said genetic modification further comprises incorporation of at least one expression system for a polyketide synthase protein.

8. The cells of claim 7 wherein said genetic modification comprises incorporation of at least one expression system for a phosphopantetheinyl transferase.

9. The cells of claim 4 wherein said at least one polyketide synthase protein is derived from erythromycin, oleandomycin, megalomycin, picromycin, FK506, FK520, rapamycin, spinosad, avermectin, tylosin or epothilone.

10. A method to produce a polyketide which method comprises culturing the cells of claim 1 under conditions wherein said polyketide is produced.

11. A method to assess the results of a procedure effecting modification of polyketide synthase genes, resulting in a mixture of said modified genes which method comprises

transfecting a culture of *E. coli* of claim 3 with said mixture of modified genes,  
culturing individual colonies of said transformed *E. coli*, and  
assessing each colony for polyketide production

12. The method of claim 11 wherein said *E. coli* have been modified to contain a functional phosphopantetheinyl transferase, a functional propionyl CoA carboxylase and have further been modified to delete the *prpA-D* operon.

13. A method to enhance the production of a polyketide in a microbial host which method comprises providing said host with an expression system for a first enzyme that catalyzes the production of starter and/or extender units used in constructing the polyketide.

14. The method of claim 13 wherein said first enzyme is propionyl CoA carboxylase.

15. The method of claim 14 wherein said propionyl CoA carboxylase is encoded by the *pccB* and *accA2* genes from *S. coelicolor*.

16. The method of claim 13 wherein said first enzyme is malonyl CoA decarboxylase.

17. The method of claim 16 wherein the malonyl CoA decarboxylase is encoded by the *matA* gene from *R. trifoli*.

18. The method of claim 13 wherein said first enzyme is malonyl CoA synthetase.

19. The method of claim 18 wherein the malonyl CoA synthetase is encoded by the *matB* gene of *R. trifoli*.

20. The method of claim 48 which further includes providing the substrate for malonyl CoA synthetase and an expression system for a second enzyme that effects entry of said substrate into the cell.

21. The method of claim 20 wherein the second enzyme is encoded by the *matC* gene of *R. trifoli*.

22. The method of claim 20 wherein said substrate is of the formula  $R_2C(COOH)_2$  wherein each R is H or is an optionally substituted hydrocarbyl group of 1-8C.

23. The method of claim 22 wherein one R is H, methyl or ethyl and the other is H.

24. Recombinant microbial cells that produce at least one polyketide which cells have been modified to contain an expression system for a nucleotide sequence encoding at least one enzyme that enhances the production of a starter and/or extender unit of said polyketide.

25. The cells of claim 24 which are *Streptomyces* or *Escherichia*.

26. The cells of claim 25 which are *Streptomyces coelicolor* CH999 or *E. coli*.

27. The cells of claim 26 which are *E. coli*

28. The cells of claim 27 wherein said polyketide is a complete polyketide.

29. The cells of claim 28 wherein said polyketide is 6-dEB.

30. A method to produce a polyketide which method comprises culturing the cells of claim 24 under conditions wherein said polyketide is produced.

31. The method of claim 30 wherein precursor for starter and/or extender is added to the medium.

5 32. The method of claim 31 wherein said at least one precursor is a diketide.

33. A reaction mixture for the production of a polyketide which reaction mixture comprises, in addition to enzymes catalyzing the production of said polyketide, at least one enzyme which catalyzes the conversion of a substrate to an extender or starter unit for said polyketide.

10 34. The reaction mixture of claim 33 wherein said first enzyme is propionyl CoA carboxylase.

35. The reaction mixture of claim 34 wherein said propionyl CoA carboxylase is encoded by the *pccB* and *accA2* genes from *S. coelicolor*.

15 36. The reaction mixture of claim 33 wherein said first enzyme is malonyl CoA decarboxylase.

37. The reaction mixture of claim 36 wherein the malonyl CoA decarboxylase is encoded by the *matA* gene from *R. trifoli*.

38. The reaction mixture of claim 36 which further includes providing the substrate for malonyl CoA synthetase and a substrate therefor.

20 39. The reaction mixture of claim 37 wherein said substrate is of the formula  $R_2C(COOH)_2$  wherein each R is H or is an optionally substituted hydrocarbonyl group of 1-8C.

43. The cells of claim 42 wherein the polyketide is 6-dEB.

add B1      add C10

$\frac{d}{dt} \left( \frac{\partial L}{\partial \dot{x}} \right) = \frac{\partial L}{\partial x}$